"Synergistic Improvements to Polynucleotide Vaccines" 09/828,505 Eyal Raz, et al.



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FIG. 1

Construct of Amb a1 cDNA - II

Full-length Amb a1 (396 codon)

Plant Leader Sequence (36AA)

\$\Delta 36 \text{ Amb a1}\$

\$\ssha 36 \text{ Amb a1}\$

Virus Leader Sequence (14AA)

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FIG. 2

Comparison of codon usage (Plant vs. Human)

71	T	C
П	1	O

	Plant	Human
CAT	83%	0%
CAC	17%	100%

GLN

	Plant	Human		
CAA	90%	30%		
CAG	10%	70%		

ASP

	Plant	Human
GAT	76%	31%
GAC	24%	69%

GLU

	Plant	Human
GAA	69%	25%
GAG	31%	75%

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FIG. 3

Expression of Amb a1 in COS-7 cell - III

43 kDa	1	2	3	4	5	•	
43 KDa_						-39	kDa
38 kDa [√]	The second second			2 ,731 \			

1: Purified AgE

Tr. 27 Labor 17 Labor

2: Amb a1/pNDKm (x1)

3: \triangle 36Amb a1/pNDKm (x3)

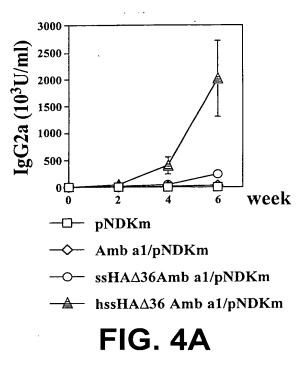
4: $ssHA\Delta 36Amb \ a1/pNDKm$ (x 3)

5: $hssHA\Delta 36Amb \ a1/pNDKm \ (x 10)$

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Induction of Antigen-specific Antibody and Cytokine in vivo - III

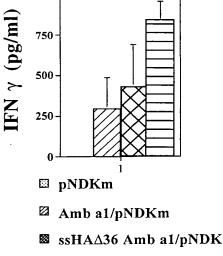
1000



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- S ssHA∆36 Amb a1/pNDKm
- **■** hssHA∆36 Amb a1/pNDKm

FIG. 4B

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Induction of Antigen-specific Antibody and Cytokine in vivo - VI

(Co-injection of ISS-ODN with $50\mu g$ of hssHA $\!\Delta 36Amb$ a1/pNDKm)

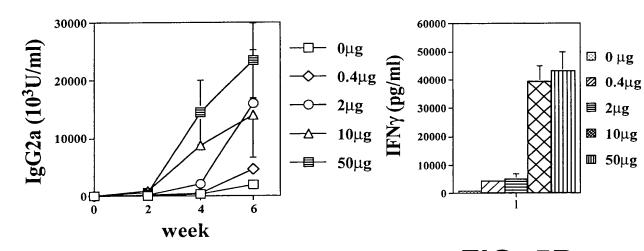


FIG. 5A

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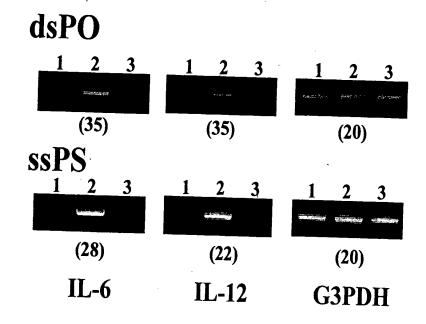
FIG. 5B

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FIG. 6

In vivo Efficacy of ISS-ODN (dsPO vs. ssPS)



1: PBS, 2: ISS-ODN, 3: M-ODN

- Injection of ISS-ODN(i.v.) [200µg/mouse]
- Isolation of spleen after 2 hr post-injection
- Isolation of mRNA
- Detection of IL-6/IL-12 transcript by RT-PCR

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10µg/mouse 0.5 mg/mouse

50µg/mouse

Reduction of Amb a1-specific IgE in vivo (week 8)

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FIG. 7A

